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The behavior of the pollen-tube in Houstonia coerulea

Chester Arthur Mathewson

The present study was undertaken at the instance of Professor F. E. Lloyd during the past winter, and prosecuted in the laboratory of Teachers College with his assistance and under his direction. My work, the material for which was collected in the neighborhood of Mt. Holyoke, Mass., in the summer of 1904, has thus far been confined to one species, *Houstonia coerulea.*

Methods

Both young and old flowers were collected and fixed in Flemming's fluid, chromo-acetic solution, and alcohol-acetic solution. With the tissue that had been fixed in the first two I used the Flemming triple stain. The tissue fixed in alcohol-acetic solution was stained with iron-alum haematoxylin and Bismarck brown. The latter method has thus far yielded the best results. All the sections were cut 10 μ thick.

Results obtained from allied forms

In connection with his work on the embryology of the *Rubiaceae*, Lloyd has studied the pollen-tube in two genera — *Richardsonia* and *Diodia.* Briefly stated, his results show that in *Richardsonia pilosa* and in *Diodia teres* the course of the pollen-tube is chiefly intercellular, while in *D. virginiana* the pollen-tube grows freely in the cavity of the ovary. Since the behavior of the tube is so different in two closely related species of the same genus, and likewise in two closely related genera of the same family, Lloyd agrees with Murbeck that the phenomenon is a matter of physiological meaning only, and disagrees with Treub and Nawaschin in their contention that the intercellular mode of growth is of importance from a phylogenetic point of view. It will be remembered in this connection that Longo, as a result of his studies in the Cucurbitaceae, has come to conclusions similar to those of Lloyd. I may add that my own results seem to add further evidence in support of this view.

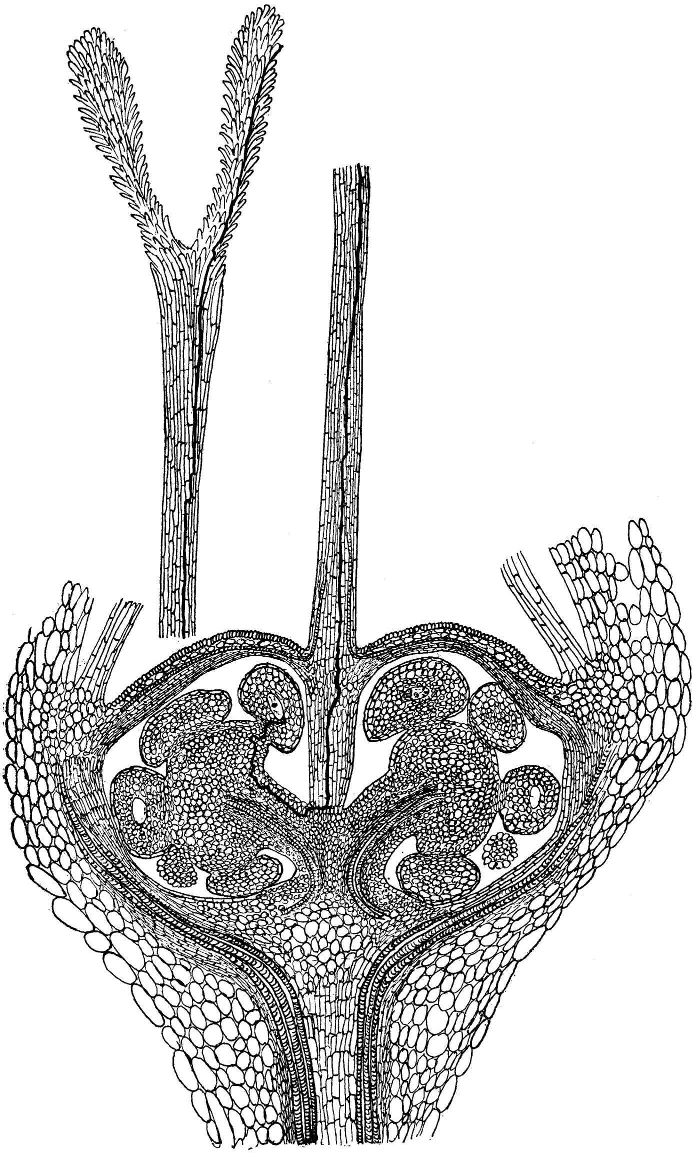

FIG. 1. Longitudinal section of the pistil of *Houstonea coerulea*, showing the course of the pollen-tube from the stigma to an embryo-sac, at its egg pole.

HISTOLOGY

In *Houstonia coerulea* there is a bilocular inferior ovary 1.9 mm. wide in its longer axis, and about 1.3 mm. deep (figure 1). The locules, as in the case of *Diodia* and *Richardsonia*, are separated by a partition of two portions. The lower portion arises, as Lloyd has shown, from the floor of the ovary. The original single ridge gives rise to two club-shaped placentae, one in each locule. Each placenta soon begins to show a number of protuberances — the primordia of the ovules. The style is single, with two stigmas, and arises by the concrescence of two ridge-like fundaments which develop laterally and opposite to each other on the inside of the hollow torus. During this concrescence a transverse ridge is formed on the inside of the ovary roof which by growing downward until it meets the ridge arising from the floor, ultimately divides the originally single ovarian chamber into two locules. The main point concerning the ovule is that there is no integument whatever at any stage, and as a result the micropyle is entirely wanting. The ovules, usually at least 15 in each locule, are very small, averaging about 70 μ long by 60 μ deep. They are when mature somewhat bowl-shaped with the concave side toward the placenta. The funicle is very short. The ovule conforms, broadly speaking, to the campylotropous type. The embryo-sac lies buried about midway in the ovule with its slightly curved longer axis at right angles with the funicle, coincident with the longer axis of the ovule.

BEHAVIOR OF THE POLLEN-TUBE

The two stigmas are furnished with numerous club-shaped papillae (figure 1), which are found over their entire surfaces. These papillae are formed by the outward bending of superficial cells. The pollen-grains average 15 to 17 μ in diameter, and are about twice the diameter of the papillae. They have numerous germ-pores.

As a rule the tube emerges from the pollen-grain on the side toward the papilla, but there are many exceptions to this. In some cases the pollen-tubes have been observed to emerge through a germ-spot diametrically opposite to the point of contact with the stigma, growing down upon the surface of the grain and so reaching the hair. In a similar case the tube grew out toward the adjacent papilla and pursued its way thereon instead of on the one with which the grain was in contact. Strasburger figures cases in *Agrostemma* and *Malva* in which the tube enters the papillary cell and pursues its way within, but I have found no evidence that this occurs in the form I have studied. Growth on the papilla is entirely superficial. The tube very often makes one, or even two full turns around a papilla before it reaches the base, a condition precisely like that described by Strasburger in the case of one of the grasses, *Alopecurus pratensis.*

Arrived at the base of the papilla the tube begins its intercellular course. The papillae merge at their bases with long, narrow relatively thin-walled cells, the first kind of conductive tissue which it meets in its course. There are no intercellular spaces whatever in the tissues traversed by the tube. The width of the pollen-tube varies from 2 to 4 μ. It seems quite probable that the method by which the tube first penetrates between the cells of the epidermis of the stigma is the method by which it is able to force its way throughout its entire course. It is well known that the pollen-tube is capable of secreting a cellulose-dissolving enzyme, and it seems that we should be safe in saying that by breaking down the middle lamella between the cells with which it comes in contact the slender tube of this species makes room for its growth. The tube is doubtless nourished, in part at least, by the absorption of the cellulose which has been converted into available food by its enzymes. The width of the tube is not constant, usually being somewhat greater in its younger portion than farther back, where it becomes collapsed. The vascular elements of the style are in two small bundles which are always farther from the middle of the style than the pollen-tubes. Little or no distortion or other bad effect results from the contact of the pollen-tube with the cells along its course. If the tube meets a cell lying directly across its path it simply passes around it.

At the level of the roof of the ovary the pollen-tube encounters a tissue, the cells of which are more irregular and the walls thicker (figure 1). Some of the cells are quite short. The tube goes on down through this stylar portion of the ovarian partition as readily as through the part above. Anatomically there appears no reason why it should not make its way into the cavity of the ovary directly after reaching this tissue, but it appears that it never does so.

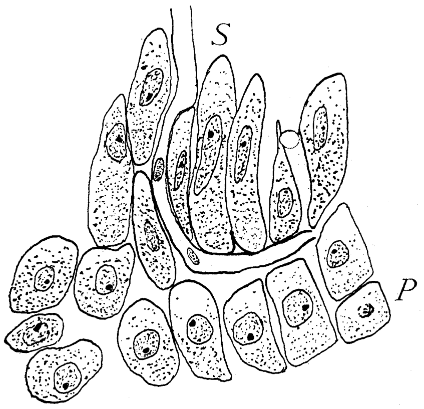

FIG. 2. Contact of stylar tissue (*S*) with the basal element of the ovary-partition (*P*), showing the turn made by the pollen-tube on reaching the latter.

At the place where the stylar elements blend with the basal elements of the partition the tube encounters a third kind of tissue which is in sharp contrast with those heretofore traversed. Its course appears to be interrupted by relatively shorter, columnar cells with thicker walls beneath which are parenchymatous cells. On reaching this tissue the tube invariably makes a sharp turn to the right or left (figure 2), and thus enters the stalk of one of the two placentae. It often happens that this turn takes the tube directly toward the surface of the placenta, and in such a case the tube makes a further turn which carries it beneath the epidermis (figure 3). In no case does it approach nearer than within one cell-layer of the surface. Large cellulose plugs (figure 3), twice the diameter of the pollen-tube itself, are often seen in this region. They also occur quite often in the style.

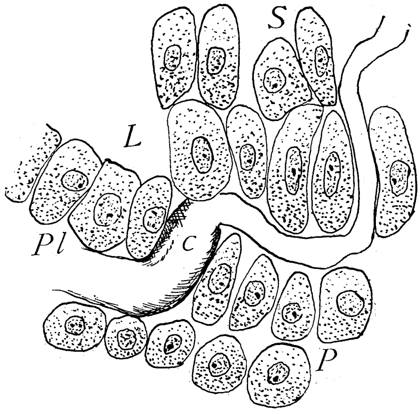

FIG. 3. Basal portion of placental stalk (*Pl*) and adjacent tissue (*P*) of the basal partition, with a part of the stylar tissue (*S*) abutting on the latter. *C*, cellulose plug in the pollen-tube, which at this point turns in order to penetrate the placental stalk. *L*, locule.

The pollen-tube continues its course through the placenta either immediately below the epidermis or from one to several cell-rows below it. This tissue is composed of approximately isodiametric cells and the path of the tube in it may be straight or subject to sudden variations. As the tube approaches the ovule it usually grows toward the interior of the placenta, so that by the time it reaches the region of the funicle, it may be six cell-rows below the surface. The cells of the funicle constituting a scission layer, are much smaller than the parenchymatous cells of the placenta and have their long axes at right angles to the path of the tube. The tube usually makes many turns in passing through this tissue. It may even grow for a minute distance across the sinus between the funicle and the ovule.

The tube has now reached the tissue of the ovule, composed of small thin-walled cells, and its course, as in *Alchemilla* (Murbeck), in the region of the egg is subject to sudden deviations. The general direction, however, is toward the egg end of the embryo-sac, which it finally reaches. The course of the pollen-tube, it will be seen, is very closely analogous to that in *Alchemilla* (Murbeck), in the *Cannabinaceae* (Zinger), in *Cynomorium* (Pirotta and Longo), and in *Ulmus* (in some instances studied by Nawaschin).

SUMMARY AND CONCLUSIONS

The cells with which the pollen-tube comes in contact influence it only in a passive manner.

The influence of the pollen-tube upon the cells with which it comes in contact is very slight.

The course of the tube seems to support the hypothesis that has been advanced that a chemotactic stimulus originating in the egg-apparatus, or the egg itself, is the chief directive influence.

The endotropic phenomenon here set forth adds another link in the chain of evidence that the intercellular mode of growth is purely a physiological phenomenon, and not a character inherited from more primitive forms.

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